K101185

Iris Molecular Diagnostics

A Division of IRIS International, Inc.

SEP 20 2011

510(k) Summary
NADiA® ProsVue™

This summary of 510(k) safety and effectiveness is being submitted in accordance with the requirements of SMDA 1990 and 21 CFR 807.92.

1. **Submitter name and address contact**

Iris Molecular Diagnostics
2075 Corte del Nogal, Suite J
Carlsbad, CA 92011
Telephone: (760) 795-5003
Fax: (760) 438-9943
Contact Person: Robert E. Klem, PhD
Date Prepared: September 19, 2011

2. **Device Name**

Proprietary Name: NADiA® ProsVue™
Common Name: Immuno-PCR Assay for the Determination of Prostate Specific Antigen (PSA)
Classification Name: Prostate-specific antigen (PSA) for prognostic, recurrence risk assessment of prostate cancers

3. **Predicate Device**

Agendia BV MammaPrint® (k062694)

4. **Device Description**

ProsVue™ is a two-site immunoassay utilizing an assay specific synthetic DNA sequence as a label with a PCR detection method. Calibrators, controls and samples react with a reagent containing a monoclonal PSA-specific antibody labeled with an assay-specific double-stranded DNA sequence. Then a reagent of paramagnetic microparticles coupled to a monoclonal antibody specific for another site on PSA is added and allowed to react to form a specific sandwich complex with PSA. After washing the particles to remove reactants, a reagent containing a heat-stable polymerase, specific primers, nucleotides, and a fluorescent dye is added to the washed microparticles. An Applied Biosystems® (AB) 7500 Fast Dx Real-Time PCR instrument is utilized to detect the presence of the monoclonal PSA-specific antibody labeled with an assay specific DNA sequence indicating the levels of PSA in the samples. PSA values of controls and samples are calculated in pg/mL from a calibrator dose-response plot. ProsVue slope is calculated using ProsVue Software from the

calculated PSA concentrations of three patient samples collected between six weeks and 20 months post radical prostatectomy.

The assay is made up of Reporter Antibody Reagent, Target Capture Reagent, PCR Reagent, Calibrators, Directions for Use (DFU) and ProsVue Software. Wash Reagent, Sample Diluent, and a three-level assayed control set are provided separately. ProsVue users initially receive the DFU and ProsVue software, which provide instructions and quality control support for the process of sample scheduling, collection and storage. When samples are ready for testing, the user contacts Iris Molecular Diagnostics (IMD), and IMD ships the required reagents.

5. Intended Use

NADiA® ProsVue™ is an in-vitro diagnostic assay for determining rate of change of serum total prostate specific antigen over a period of time (slope, pg/mL per month). The NADiA® ProsVue™ assay is performed for patients having less than 0.1 ng/mL serum total PSA values (determined by standard-of-care assays that are FDA approved/cleared) in the first sample collected more than 6 weeks after radical prostatectomy. ProsVue™ slope is indicated for use as a prognostic marker in conjunction with clinical evaluation as an aid in identifying those patients at reduced risk for recurrence of prostate cancer for the eight year period following prostatectomy.

The NADiA® ProsVue™ assay is not intended for the diagnosis or for the monitoring of prostate cancer.

6. Assay Performance

Measuring Interval

From 0.65 pg/mL to 100 pg/mL

Sensitivity

Limit of blank, limit of detection, and limit of quantitation were determined with the Clinical Laboratory Standards Institute (CLSI) EP17-A guideline.

Limit of Blank (LOB):	0.17 pg/mL
Limit of Detection (LOD):	0.27 pg/mL
Limit of Quantitation (LOQ):	0.65 pg/mL

Linearity

Linearity was evaluated in accordance with the CLSI EP06-A guideline. For total PSA by NADiA® ProsVue™, the method has been demonstrated to be linear from 0.65 pg/mL to 100 pg/mL with deviation from linearity less than 24%.

Precision

Three male serum pools were analyzed in accordance with CLSI EP5-A2. Samples were analyzed in duplicate determinations at two sites, by three operators using two AB 7500 Fast Dx instruments and two reagent lots. Forty assays were performed at the first site by two operators. Each operator performed one run per day, alternating between reagent lots, over a course of 20 days. At the second site, a single operator performed 2 runs a day, one on each reagent lot, for 5 days. The two sites combined yielded a total of 50 assays over a period of 25 non-consecutive days. Two statistical analyses were performed to analyze contribution of components of variation to total variation: one for estimation of within-run, between-run, and between-day components of variation; and a second for estimation of within-run, between-run, and between-lot components of variation. Additionally, site-specific results were calculated. The following results were obtained:

A. Within-run, between-run, and between-day variation

Measure	Low Sample	Intermediate Sample	High Sample
Mean (pg/mL)	3.79	24.1	69.1
Within-run variation			
SD	0.34	1.74	5.97
%CV	9.0	7.2	8.6
Between-run			
SD	0.34	0.81	3.22
%CV	9.0	3.4	4.7
Between-day			
SD	0.32	1.23	2.83
%CV	8.3	5.1	4.1
Total variation			
SD	0.58	2.28	7.35
%CV	15.2	9.4	10.6

Components of variation estimated only include within-run, between-run, and between-day in this analysis.

B. Within-run, between-run, and between-lot variation

Measure	Low Sample	Intermediate Sample	High Sample
Mean (pg/mL)	3.79	24.1	69.1
Within-run variation			
SD	0.21	0.42	1.35
%CV	5.6	1.7	2.0
Between-run			
SD	0.47	1.48	4.37
%CV	12.5	6.1	6.3
Between-lot			
SD	0.15	0.28	0.00
%CV	4.0	1.1	0.0
Total variation			
SD	0.75	1.71	4.57
%CV	14.0	6.5	6.6

Components of variation estimated only include within-run, between-run, and between-lot in this analysis.

The between-lot imprecision was not more than 4.0%.

C. Site-specific results

Site(s)	Days	Data points	Sample #	Mean (pg/mL)	Between-site %CV
1	20	80	1	3.96	
			2	24.4	
			3	70.7	
2	5	20	1	3.11	
			2	23.0	
			3	62.5	
1 & 2	5[1] + 5	40	1	3.55	13.7
			2	24.2	7.3
			3	66.9	9.6

[1] 5 + 5 = first 5 days at site 1 and entire 5 days at site 2

Interfering Substances
Elevated concentrations of blood constituents and drugs were added to serum samples containing PSA at 3 and 50 pg/mL and assayed in quadruplicate with the ProsVue™ assay. The substances added and the highest concentrations tested are listed in the tables below. At the concentrations listed, these substances showed less than 10% interference in assay results.

Interference by blood constituents	
Bilirubin, conjugated	30 mg/dL
Cholesterol	500 mg/dL
Creatinine	5.0 mg/dL
Hemoglobin	200 mg/dL
Immunoglobulin G	6 g/dL
Triglycerides	1000 mg/dL
Urea	260 mg/dL
Uric acid	23.5 mg/dL

Interference by drugs	
10-hydroxynortriptyline	700 ng/mL
5-Fluorouracil	390 µg/mL
Acetaminophen	200 µg/mL
Ampicillin	53 µg/mL
Ascorbic acid	60 µg/mL
Biotin	50 ng/mL
Caffeine	60 µg/mL
Carbamazepine	30 µg/mL
Chloramphenicol	50 µg/mL
Cimetidine	20 µg/mL
Ciprofloxacin	10 µg/mL
Cisplatin	12 µg/mL
Cotinine	1.9 µg/mL
Cyclophosphamide	375 µg/mL
Dextran-40	60 mg/mL
Digoxin	6.1 ng/mL
Doxorubicin	240 ng/mL
Erythromycin	60 µg/mL

Ethanol	4 mg/mL
Ethosuximide	250 µg/mL
Flutamide	500 ng/mL
Furosemide	60 µg/mL
Gentamicin	10 µg/mL
Heparin sodium	3 U/mL
Ibuprofen	500 µg/mL
Leuprolide acetate	200 ng/mL
Lidocaine	12 µg/mL
Lithium	22.5 µg/mL
Methotrexate	910 µg/mL
Paclitaxel	6.5 µg/mL
Pamidronate	9 µg/mL
Phenytoin	50 µg/mL
Prednisone	300 ng/mL
Primidone	40 µg/mL
Prochlorperazine	1 µg/mL
Salicylic acid	600 µg/mL
Sulfamethoxazole	400 µg/mL
Tamoxifen	1.5 µg/mL
Trimethoprim	40 µg/mL
Valproate sodium	500 µg/mL
Vancomycin	100 µg/mL
Vinorelbine	1.2 µg/mL

7. Stability of PSA in serum at <100 pg/mL

To demonstrate that low-level PSA samples observed post-RP have a similar stability profile as those with values above 100 pg/mL when stored at -70 °C, serum pools from post prostatectomy patients containing PSA concentrations at ~7 and ~150 pg/mL were tested in an accelerated stability study at 4, 20, 30 and 40 °C. Aliquots of each pool were assayed with NADiA® ProsVue™ at baseline and at serial time points over 15 days at each incubation temperature. Stability projections indicated that 99.6% and 99.7% of PSA would remain immunoreactive at 20 years of storage at -70 °C based on the observed decomposition rates of the low and high serum pools, respectively.

8. ProsVue Reagent and Control Stability

ProsVue reagent stability was evaluated for three lots of ProsVue reagents that were stored at the labeled storage temperatures and tested at multiple time points. The kit reagents and wash solution were tested as a kit using in-house controls as test samples. The expiration period was defined as the elapsed time up to the point (the first time point of three successive time points) when the mean control recovery from three consecutive time points fell outside the value-assigned control limits.

ProsVue Sample Diluent stability was evaluated for three lots of sample diluent that were stored at the labeled storage temperature and tested at multiple time points. At each time point the Sample Diluent was used to make a 1:200 dilution of a high PSA control, and the diluted sample was tested with the ProsVue assay. The expiration period was defined as the elapsed time up to the point (the first time point of the successive time points) when the mean recovery from three consecutive time points fell outside the value-assigned range for the 1:200 dilution of the high control.

ProsVue Controls stability was evaluated for three lots of ProsVue controls that were stored at the labeled storage temperature and tested at multiple time points using the ProsVue assay. The control expiration period was defined as the elapsed time up to the point (the first time point of three successive time points) when the mean control recovery from three consecutive time points fell outside the value-assigned limits.

The results of the ProsVue stability testing are summarized in the table below. Additional stability testing will be performed, and expiration dates will be assigned based upon the stability data available. Reagents will be shipped to ProsVue users when the user notifies IMD that samples are available to be tested.

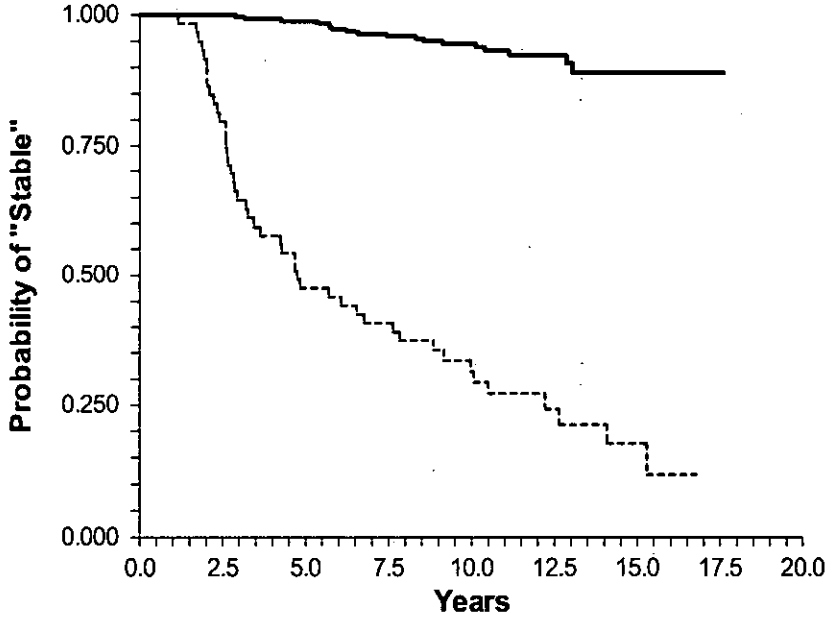
ProsVue Reagent and Control Stability

Component	Storage Conditions	Shelf Life (Days)
Kit		41
Calibrators	-30°to -10°C	41
PCR Reagent	-30°to -10°C	41
Control Level 1	-30°to -10°C	78
Control Level 2	-30°to -10°C	78
Control Level 3	-30°to -10°C	78
Target Capture Reagent	2°to 8°C	41
Reporter Antibody Reagent	2°to 8°C	41
Wash Solution	15°to 30°C	41
Sample Diluent	-30°to -10°C	41

9. Clinical Performance

To evaluate the prognostic capability of ProsVue™ linear slope to identify prostate cancer patients at reduced risk of prostate cancer recurrence following radical prostatectomy (RP), 304 patients were selected using a case-cohort study design. Patients meeting the study entry criteria were acquired from four clinical sites where each patient's clinical status ("Stable" or "Recurrence") was documented. Samples used were from men age 41 to 79 years with documented prostate cancer treated via radical prostatectomy with curative intent. Caucasian (88.8%), African-American (8.2%), Asian (2.0%) and patients of unknown race (1.0%) achieving post-RP total PSA values <100 pg/ml by standard-of-care assays were followed up to 17.6 years post-prostatectomy with three serum samples per patient acquired at various time points during the 1.5-20 month ProsVue sampling period. The study cohort consisted of 64 patients with "Recurrence" and 240 "Stable" patients. Recurrent patients were followed a median of 4.7 years (range 1.2 – 15.3 years) until "Recurrence" was documented by positive imaging, positive biopsy, or death due to prostate cancer. "Stable" patients were followed for a median of 11.0 years (range 8.0 – 17.6 years).

Figure 1: Kaplan-Meier plot of probability of stable classification versus years (Solid line indicates patients with ProsVue slope ≤ 2 pg/ml/month [n=245] and dashed line indicates patients with ProsVue slope > 2 pg/ml/month [n=59]).



The Kaplan-Meier estimates of the 8-year probability of "Stable" were 95.9% (95% CI: 93.4 - 98.4%) for the patients with ProsVue Slope ≤ 2

pg/mL/month and 37.3% (95% CI: 25.0 - 49.6%) for the patients with ProsVue Slope > 2 pg/mL/month. Median time to recurrence in the patients with ProsVue slope ≤ 2 pg/ml/month was > 17.6 years versus 4.8 years for patients with ProsVue slope > 2 pg/ml/month.

Further investigation with Cox proportional hazards regression analysis used to evaluate the association between ProsVue linear slope and prostate cancer recurrence status in a univariate analysis (can be interpreted as stand alone performance) indicated that the patients with ProsVue Slope≤2 pg/mL/month were 18.3 times more likely to be "Stable" than those with ProsVue Slope>2 pg/mL/month (HR = 18.3 with 95% CI: 10.6 to 31.8; p<0.0001). Thus, ProsVue linear slope was a significant predictor of reduced risk for prostate cancer recurrence in this analysis (Table 1).

Table 1: Univariate Cox proportional hazards regression results for ProsVue Slope

Term	HR	HR 95% CI	p-value
ProsVue linear slope (> 2.0 pg/ml/month vs. ≤ 2.0 pg/ml/month)	18.332	10.6 - 31.8	<0.0001

Cox proportional hazards regression analysis adjusted for, pre-specified covariates of pre-prostatectomy PSA value, pathologic disease stage and Gleason score was performed with the ProsVue slope term (Table 2). Of the covariates, only the Gleason score variable reached significance (p = 0.0004). The ProsVue linear slope term was attenuated from the univariate analysis but remained a highly significant and independent predictor of risk for prostate cancer recurrence with a HR of 9.8 (95% CI: 5.4 to 17.8, p<0.0001). The reduced hazard probably reflects that ProsVue slope may be related to the clinical factors but is clearly providing additional information.

Table 2: Multivariate Cox proportional hazards regression results for ProsVue™ Slope

Term	HR	HR 95% CI	p-value
ProsVue linear slope (> 2.0 pg/ml/month vs. ≤ 2.0 pg/ml/month)	9.824	5.4 - 17.8	<0.0001
Pre-prostatectomy PSA value	1.006	0.98 - 1.03	0.6469
Pathologic disease stage	1.729	0.89 - 3.4	0.1052
Gleason score	5.389	2.1 - 13.8	0.0004

The probability of clinical status "Recurrence" for the subjects with Slope>2 pg/mL/month (Positive Predictive Value, PPV) and probability to remain with clinical status "Stable" through at least 8 years for the subjects with slope ≤ 2 pg/mL/month (Negative Predictive Value, NPV) were calculated from the clinical study results (n=304). Table 3 presents a 2 x 2 table of the ProsVue classification based on the slope outcome and to the clinical status of patients as "Stable" or "Recurrence" following 8 years (row and column totals are displayed).

Table 3: 2x2 Table of ProsVue™ classification vs. Reference Clinical status (N=304)

		Reference Clinical Status		
		Recurrence	Stable	Totals
ProsVue	*Slope > 2 pg/mL/month*	46	13	59
	Slope ≤ 2 pg/mL/month	18	227	245
	Totals	64	240	304

Among subjects who had "Recurrence" during 8 years of follow-up, 71.9% (46/64) subjects had slope > 2 pg/mL/month and among the subjects who had "Stable" through at least 8 years of follow-up, 94.6% (227/240) subjects had slope ≤ 2 pg/mL/month.

Positive likelihood ratio (PLR) was 13.3 with 95% CI: 7.7 – 23.0 and negative likelihood ratio (NLR) was 0.297 with 95% CI: 0.201 – 0.440.

In these data, probability of "Recurrence" regardless of the values of the ProsVue slope was 21.0% (64/304); PPV was 78.0% with 95% CI: 65.3 – 87.7% and NPV was 92.7% with 95% CI: 88.6 – 95.6%.

Because the estimates of PPV and NPV depend on the probability of "Recurrence" among all subjects (prevalence of clinical status "Recurrence"), Table 4 presents estimates of PPV and NPV calculated for various levels of prevalence of "Recurrence".

Table 4: Estimates of PPV and NPV calculated for various levels of prevalence (%) of clinical recurrence.

Prevalence (%)	PPV (%)	NPV (%)
10	59.7	96.8
15	70.1	95
20	76.9	93.1
25	81.6	91.0
30	85.1	88.7
35	87.8	86.2

A subgroup analysis was performed on data from 20 patients with at least one post-RP PSA value ≥ 300 pg/mL, but who remained stable throughout at least 8 years of follow-up. ProsVue slope correctly classified 11 of the 20 patients as "at reduced risk for recurrence" (ProsVue slope ≤ 2 pg/mL/month).

9. Software

Software is provided as an accessory to the ProsVue™ assay kit. Its purpose is to reduce the risk of errors in the use of appropriate serum samples and calculating assay results.

The ProsVue™ software identifies serum samples with invalid collection intervals and calculates the equation of the calibration line, PSA concentration (pg/mL) of patient samples and assay controls, patient ProsVue slope (pg/mL/month), and patient risk category. ProsVue software generates a report that includes the ProsVue slope (in pg/mL/month) and whether that slope categorizes a patient as being at "reduced risk" or "not at reduced risk" of prostate cancer recurrence.

ProsVue™ software is an Excel Add-In containing calculation spreadsheets, Userforms, and code modules. Its User Interface consists of Userforms and dialog boxes consistent with those found in standard Excel Add-Ins. The User Interface allows entry and validation of sample identifiers and dates, plate map set-up, selection of a raw data (.csv) file to be analyzed, and viewing/printing of assay results. All sample and assay run requirements, as provided in the ProsVue™ Directions for Use are applied in the software to ensure valid results.

Conclusion

NADiA® ProsVue™ is substantially equivalent to the predicate device, the Agendia BV MammaPrint® (k062694). No new issues of safety or effectiveness have been raised.

DEPARTMENT OF HEALTH & HUMAN SERVICES

Public Health Service

Food and Drug Administration
10903 New Hampshire Avenue
Silver Spring, MD 20993

Iris Molecular Diagnostics
c/o Robert E. Klem, Ph.D.
Vice President of Product Development
2075 Corte Del Nogal, Suite-J
Carlsbad, CA 92011

SEP 2 0 2011

Re: k101185
Trade/Device Name: NADiA® ProsVue™
Regulation Number: 21 CFR 866.6040
Regulation Name: Gene expression profiling test system for breast cancer prognosis
Regulatory Class: II
Product Code: OWM
Dated: September 5, 2011
Received: September 7, 2011

Dear Dr. Klem:

We have reviewed your Section 510(k) premarket notification of intent to market the device referenced above and have determined the device is substantially equivalent (for the indications for use stated in the enclosure) to legally marketed predicate devices marketed in interstate commerce prior to May 28, 1976, the enactment date of the Medical Device Amendments, or to devices that have been reclassified in accordance with the provisions of the Federal Food, Drug, and Cosmetic Act (Act) that do not require approval of a premarket approval application (PMA). You may, therefore, market the device, subject to the general controls provisions of the Act. The general controls provisions of the Act include requirements for annual registration, listing of devices, good manufacturing practice, labeling, and prohibitions against misbranding and adulteration.

If your device is classified (see above) into class II (Special Controls), it may be subject to such additional controls. Existing major regulations affecting your device can be found in Title 21, Code of Federal Regulations (CFR), Parts 800 to 895. In addition, FDA may publish further announcements concerning your device in the Federal Register.

Please be advised that FDA's issuance of a substantial equivalence determination does not mean that FDA has made a determination that your device complies with other requirements of the Act or any Federal statutes and regulations administered by other Federal agencies. You must

comply with all the Act's requirements, including, but not limited to: registration and listing (21 CFR Part 807); labeling (21 CFR Parts 801 and 809); medical device reporting (reporting of medical device-related adverse events) (21 CFR 803); and good manufacturing practice requirements as set forth in the quality systems (QS) regulation (21 CFR Part 820). This letter will allow you to begin marketing your device as described in your Section 510(k) premarket notification. The FDA finding of substantial equivalence of your device to a legally marketed predicate device results in a classification for your device and thus, permits your device to proceed to the market.

If you desire specific advice for your device on our labeling regulation (21 CFR Parts 801 and 809), please contact the Office of *In Vitro* Diagnostic Device Evaluation and Safety at (301) 796-5450. Also, please note the regulation entitled, "Misbranding by reference to premarket notification" (21 CFR Part 807.97). For questions regarding the reporting of adverse events under the MDR regulation (21 CFR Part 803), please go to http://www.fda.gov/MedicalDevices/Safety/ReportaProblem/default.htm for the CDRH's Office of Surveillance and Biometrics/Division of Postmarket Surveillance.

You may obtain other general information on your responsibilities under the Act from the Division of Small Manufacturers, International and Consumer Assistance at its toll-free number (800) 638-2041 or (301) 796-7100 or at its Internet address http://www.fda.gov/cdrh/industry/support/index.html.

Sincerely yours,

Maria M. Chan

Maria M. Chan, Ph.D.
Director
Division of Immunology and Hematology Devices
Office of *In Vitro* Diagnostic Device
Evaluation and Safety
Center for Devices and Radiological Health

Enclosure

Indications for Use

510(k) Number (if known): K101185

Device Name: NADiA® ProsVue™

Indications for Use:

NADiA® ProsVue™ is an in-vitro diagnostic assay for determining rate of change of serum total prostate specific antigen over a period of time (slope, pg/mL per month). The NADiA® ProsVue™ assay is performed for patients having less than 0.1 ng/mL serum total PSA values (determined by standard-of-care assays that are FDA approved/cleared) in the first sample collected more than 6 weeks after radical prostatectomy. ProsVue™ slope is indicated for use as a prognostic marker in conjunction with clinical evaluation as an aid in identifying those patients at reduced risk for recurrence of prostate cancer for the eight year period following prostatectomy.

The NADiA® ProsVue™ assay is not intended for the diagnosis or for the monitoring of prostate cancer.

Prescription Use) ___X___ (Part 21 CFR 801 Subpart D) AND/OR Over-The-Counter Use _____ (21 CFR 801 Subpart C)

(PLEASE DO NOT WRITE BELOW THIS LINE – CONTINUE ON ANOTHER PAGE IF NEEDED)

Concurrence of CDRH, Office of Device Evaluation (ODE)

Reena Philip

Division Sign-Off

Office of In Vitro Diagnostic Device Evaluation and Safety

510K k101185